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Enhancing electrical communication through reconstructed nanowire electrodes for implantable enzymatic biofuel cell (EBFC)

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Abstract: Enzymatic biofuel cell (EBFC) is a promising continuous energy supply powered by abundant and essential components in human physiological fluid; glucose and oxygen, for powering implanted device. Important aspects in BFC are establishing efficient electron transfer pathways between enzyme redox center and electrodes and maintaining enzyme stability. In this study we characterized of electron transport from enzyme as core of BFC to the nanowire by edge-immobilization (EI) and sidewall-immobilization (SI). Through immobilization in both aspects of nanowire surface, electrical transport through edge is responsible for efficiency meanwhile sidewall immobilization benefits for enzyme stacking compartment. We also studied the functional group exist in nanowire may act as bridge to individual nanowire. Nanowire which is lacking of EWG diminished electron transport enhancement between C-C bonds through resistance barrier. Further study which factor limiting and enhancing electrical transport through pristine and constructed carbon nanotube is still being assessed.

Keywords: Implantable EBFC, carbon nanotube network, electrical communication, reconstructed nanowire

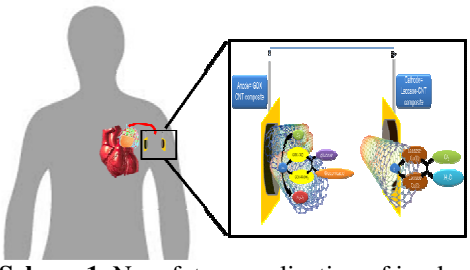
1 INTRODUCTION

Enzymatic biofuel cell (EBFC) is a system which generates electrical power from conversion of substrate through redov reaction in the enzyme redox center ((Mano, Mao et al. 2003; Soukharev, Mano et al. 2004; Cracknell, Vincent et al. 2008; Moehlenbrock and Minteer 2008). Being able to generate micropower, EBFC has been drawing out the attention for real application and developed the power source for implanted devices (MacVittie, Halamek et al. 2012) by using human essential compound; glucose and oxygen. As near future promising power source, EBFC is expected to be able to power developed implanted devices such as pacemakers, neuron stimulator and implanted neurophysiological monitoring devices (Justin, Yingze et al. 2004; MacVittie, Halamek et al. 2012).

Before Heller and Cosnier's group conducting the real application of implanted enzymatic in plant and animal, EBFC was less attractive due to its low voltage which thermodynamically limited by the potential of enzymes substrate (MacVittie, Halamek et al. 2012). Based on the EBFC main limitation, many studies had been conducted to overcome the critical points of EBFC which is the communication between enzyme and electrode (Ferreira, Iost et al. 2012). Achieving the high current and power density of EBFC is the main objective of the electrodes construction.

Conductive nanowires have been studied as a matrix for electrode architecture of EBFC. Nanowire such as carbon nanotubes (CNTs) are able to act as enhancement factor of electrical communication between enzyme and electrode superficial (Ferreira, Iost et al. 2012) and also as enzyme stacking compartment due to its large surface area. However, like other nanoparticle characteristic, CNTs also carry concerning drawback as electrode matrix where it is accompanied by high charge transfer resistance (Wang 2006) and electrolyte diffusion limitation due to its hydrophobicity. Hence, optimization of nanowire condition is practically needed to enhance electron transfer efficiency.

In this study we assessed the electrical network through CNTs in the EBFC system. Nanowire network are built in the edge-edge (E-E), edge-sidewall (E-S) and sidewall-sidewall (S-S) to support EBFC performance. Utilization of pristine carbon nanotube (CNTs) with its native high electrical conductivity needs to be improved to achieve maximal use of nanowire network through reconstructed electrical network with chemical functionalization. Acid treatment creates defect sites for activating –COOH functional groups in carbon nanotube to enhance electron transport from and to individual nanowire as –COOH is an electron withdrawing group (EWG). We also assessed the characteristic of electron transport from enzyme (glucose oxidase, GOX) as core of BFC to the nanowire as supporting material to avoid ambiguous interpretation of over-potential occurrence during electron transport.



Scheme1. Near future application of implanted BFC in human powering implanted medical devices

2. MATERIAL AND METHOD

2.1. Material

Glucose oxidase from *Aspergillus niger (type VII-S, 192 U/mg).* Laccase from *Trametes versicolor (13.6 U/mg), β*-D Glucose, 1-pyrenebutanoic succinimidyl ester, N-(-3-dimethylaminoprophyl) N'-ethylcarbodiimide, 2-thioethanol, cystamine sodium diphosphate and disodium phosphate and Si/Au wafer as substrate from Sigma Aldrich. Pristine, purified and acid treated multi walled carbon nanotube (MWNTs) was provided from Gyeongsan National University, Gyeongsan, South Korea. Phosphate buffer solution (PBS, 100 mM, pH 7.0) was prepared as supporting electrolyte for BFC performance. D-glucose solutions (10mM) prepared at least a night before to allow mutarational equilibrium. All solutions were prepared with deionized water.

2.2. Method

Characterization of enzyme immobilization as Edge-immobilization (EI) and sidewall-immobilization (SI) were constructed by covalent bonding and utilizing heterobifunctional polymer, respectively. Each immobilization method was evaluated through XPS (Thermo Fisher Scientific, U.K) spectroscopy. Characterization of each CNT material was done by FTIR (JASCO, U.S.A), Raman spectroscopy (Andor, U.S.A) and XPS. All electrochemical measurements for EBFC performance were performed in PBS buffer at 37°C.

Electrochemical method was performed using three electrodes system (BAS-EC epsilon, U.S.A) consists of Si/Au wafer, platinum and Ag/AgCl as working, counter and reference electrode respectively. Anode (CNT-GOX) and cathode (CNT-laccase) composites were placed in one membraneless compartment biofuel cell. Stirring and aeration was done for cathode reaction during performance test with electrolyte flow 0.1mL/min. A surface area of 0.196 cm^2 used for calculation of the current and power density.

3. RESULT AND DISCUSSION

3.1. Vertically and horizontally aligned CNTs

Direct electron transport can be achieved by enzyme immobilization in the edge of CNT shown at potential of FAD as glucose oxidase redox center, which was -0.4 V (against Ag/AgCl). EI enzyme wiring achieved electron transfer with K_s value of 1620.5/s whereas SI enzyme K_s is 953.6/s. Furthermore, SI enzyme wiring contributed to the over-potential happened in the electron transport from redox center to the electrode characterized by cathodic peak at -0.3 V instead of -0.4 V (**Fig.1**).

However, SI produced 1.5 times higher current EI indicating, more load of enzyme in the sidewall than in the edge which can be confirmed through nanowire diameter-length ratio. Through immobilization in both aspects of nanowire surface, electrical transport through edge is responsible for efficiency meanwhile sidewall immobilization benefits for enzyme stacking compartment

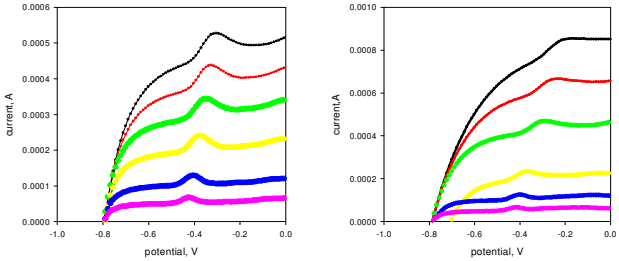


Fig. 1. Cyclic voltammograph of vertically (left) and horizontally (right) aligned CNT for GOX immobilization

3.2. XPS profile

Each immobilization method was evaluated through XPS which showed the emerge peak of C=O bonding in the 286.25 eV for horizontally aligned CNTs showing carbonyl groups mainly present in the edge of CNTs which will react with amine group of enzyme. In the other hand, we can observe increase of π-π* stacking point in the 291 eV or also called satellite peak as the result of heterobifunctional polymer use for sidewall modification

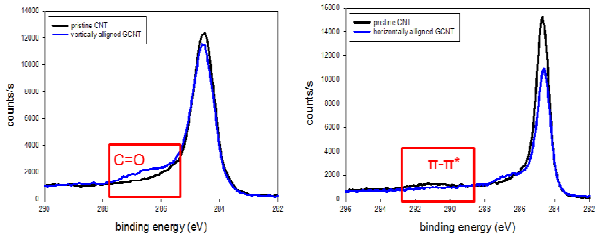


Fig. 2. XPS spectra of vertically (left) and horizontally (right) aligned CNTs on Si/Au wafer

3.3. Reconstructed nanowire with functionalization

Nanowire network are built in the edge-edge (E-E), edge-sidewall (E-S) and sidewall-sidewall (S-S) to support EBFC performance. Utilization of pristine carbon nanotube (CNTs) with its native high electrical conductivity needs to be improved to achieve maximal use of nanowire network through reconstructed electrical network with chemical functionalization.

Acid treatment creates defect sites for activating –COOH functional groups in carbon nanotube to enhance electron transport from and to individual nanowire as –COOH is an electron withdrawing group (EWG). As expected, higher current was achieved for higher degree of functionalized nanowire approximately increased 50% every 4h of strong acid treatment (**Fig. 3)**

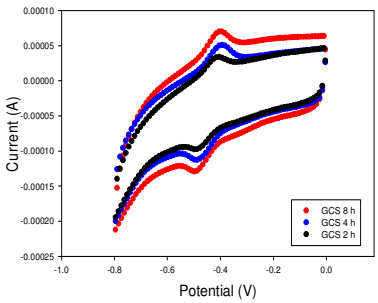


Fig. 3. Current enhancement after acid treatment

3.4. Performance of EBFC

Constructed network of CNT disk increased current and power density of BFC. By utilizing 4800 U of glucose oxidase, constructed nanowire current density was 863.00±49.33 μA/cm^2 while non-constructed network showed 67% of performance which was 583.58±49 μA/cm^2. Fig. 4. showed the performance of reconstructed and pristine CNTs-enzyme in EBFC. Reconstructed nanowire resulted 30% higher power density compared with pristine CNTs composites.

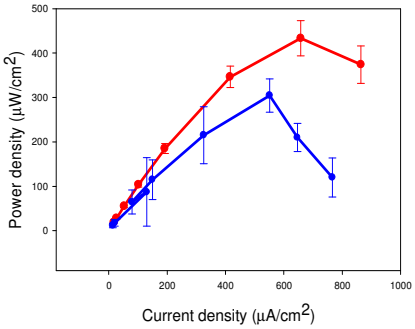


Fig. 4. I-V curve of reconstructed CNTs-GOX (red) and pristine CNTs-GOX anode composites operated in EBFC system with reconstructed CNTs-Laccase as cathode

Measurement of impedance by EIS revealed that non-functionalized CNT disk has less conductivity (3mm thick; 15,000 Ω) by higher resistance than functionalized CNT disk (3 mm thick; 9,000Ω). We suggest that functional group acts as bridge to individual nanowire which is lacking of EWG diminished electron transport enhancement between C-C bonds in CNT junction through resistance barrier. Advance assessment for the factor which limiting and enhancing electrical transport through pristine and constructed carbon nanotube is still being assessed.

4. CONCLUSION

We successfully examined the effect of enzyme stacking site to nanowire interface. Edge of CNTs is more active to enhance the electrical communication rather than sidewall. However, sidewall of CNTs is important site for enzyme stacking compartment.

Reconstructed nanowire by functionalization to make edge-like site in the sidewall will accommodate the enhancement of electrical communication by minimizing resistance between nanowire interfaces

Further factor relating to EBFC operation which affects composites performance such as electrolyte diffusion barrier and its chance to decrease the electron transfer is still being assessed.

5. REFERENCES

[1] Mano, N., F. Mao, and A. Heller, *Characteristics of a miniature compartment-less glucose-O2 biofuel cell and its operation in a living plant.* Journal of the American Chemical Society, 2003. **125**(21): p. 6588-6594.

[2] Soukharev, V., N. Mano, and A. Heller, *A four-electron O2-electroreduction biocatalyst superior to platinum and a biofuel cell operating at 0.88 V.* Journal of the American Chemical Society, 2004. **126**(27): p. 8368-8369.

[3] Moehlenbrock, M.J. and S.D. Minteer, *Extended lifetime biofuel cells.* Chemical Society Reviews, 2008. **37**(6): p. 1188-1196.

[4] Cracknell, J.A., K.A. Vincent, and F.A. Armstrong, *Enzymes as working or inspirational electrocatalysts for fuel cells and electrolysis.* ChemInform, 2008. **39**(41): p. no-no.

[5] MacVittie, K., et al., *From "Cyborg" Lobsters to a Pacemaker Powered by Implantable Biofuel Cells.* Energy & Environmental Science, 2012.

[6] Justin, G.A., et al. *Biofuel cells: a possible power source for implantable electronic devices*. in *Engineering in Medicine and Biology Society, 2004. IEMBS '04. 26th Annual International Conference of the IEEE*. 2004.

[7] Ferreira, F., et al., *Intravenous Implantable Glucose/Dioxygen Biofuel Cell with Modified Flexible Carbon Fiber Electrodes.* Lab on a Chip, 2012.

[8] Wang, P., *Nanoscale biocatalyst systems.* Current Opinion in Biotechnology, 2006. **17**(6): p. 574-579.